REMARKS/ARGUMENTS

I. Status of the Claims:

Claims 78-97, 99, 102-104, 106-109, 111-114, 116, 120-129, and 131 are pending and undergoing examination. Claims 106-109, 111, 112, 125-126, and 129 would be herein canceled without prejudice. The Applicant has previously canceled claims drawn to unelected subject matter (i.e., claims 98, 100, 101, 105, 110, 115, 117-119, and 130) without prejudice. All the pending claims would be amended. Claims 132-143 are newly presented. Upon entry of these amendments, claims 78-97, 99, 102-104, 113, 114, 116, 120-124, 127, 128, and 131-143 would be pending.

Claims 78-97, 99, 102-104, 106-109, 111-114, 116, 120-129, and 131 stand rejected for an alleged wont of enablement under 35 U.S.C §112, first paragraph.

II. The Restriction Requirement:

Applicant thanks the Examiner for reconsidering the restriction requirement and rejoining groups I, II, IV and V.

III. The Information Disclosure Statement:

Applicant submits herewith a Supplemental Information Disclosure Statement. This Statement provides additional references cited in foreign prosecution and also a Canadian, English language patent application (Reference No. AD) corresponding to EPO 437 781 B1.

IV. Status of the Specification:

The Action indicated that the application lacked an Abstract. The Abstract should be found on page 50 of the specification as filed. Applicants submit herewith a replacement page (see Appendix A), which is a true copy of the filed page and therefore presents no new matter.

V. Support for Amendments to the Claims:

The preambles of each of the pending dependent claims would be amended to replace the first occurrence of "A" by "The."

Claims 78, 99, 102-104, 123, 127, 128, and 131 would be each amended to replace each recital of "second peptidyl fragment" with the recital of "human insulin precursor peptidyl fragment." Support for this subject matter is found *inter alia* in the previous version of the claims and also in the specification in the paragraph bridging pp. 18 and 19 and at p. 19, first paragraph.

Base claims 78, 123, and 131 would be amended to recite " an arginine or lysine residue or at least one cleavable peptidyl fragment linking the first peptidyl fragment and the human insulin precursor fragment ..." Support for the subject matter of the arginine or lysine residue is found *inter alia* in the original claim 1.

Claims 102, 103, 113, 114, 127 and 128 would also be amended to recite "SEQ ID NO:" in place of "SEQ.ID.No."

Claim 78 would be amended to recite in part:

> a first peptidyl fragment of about 20 amino acids in length to 92 amino acids in length having an amino acid sequence which is at least 60% identical to the sequence of SEQ ID NO:1 at least through residues 1 - 20 of SEQ ID NO:1 and wherein the first fragment is capable of being bound by an anti-hGH antibody;
>
> a human insulin precursor peptidyl fragment comprising the human insulin A chain and the human insulin B chain; and

Support for the subject matter of "about 20 amino acids in length to 92 amino acids in length" is found in the specification *inter alia* in original claim 30, at line 14 of p. 43, and in SEQ ID NOs. 2 and 7 which teach a chaperone sequence of such lengths. Support for the subject matter of "at least 60% identical to a sequence of SEQ ID NO:1 at least through residues 1 - 20 of SEQ ID NO:1" is found *inter alia* in the previous version of the claim and in the specification at page 16, 1st full paragraph. Support for the subject matter of "wherein the first fragment is capable of being bound by an anti-hGH antibody" is found *inter alia* in original claim 3. Support for the subject matter of a "human insulin precursor peptidyl fragment comprising the human insulin A

chain and the human insulin B chain" is found in the specification *inter alia* at p. 17, third and fourth full paragraphs, and page 24, first full paragraph.

Claim 78 would also be amended to recite "from contacting" in place of "upon contact of." Support for this subject matter is found *inter alia* in the previous version of the claim.

Claim 78 would also be further amended by adding indentations for the sake of clarity.

Claim 80 depends from claim 78 and would be amended to make its recitals conform to the antecedent basis of claim 78 rather than claim 79. Support for the above is found *inter alia* in the previous version of the claim.

Claims 95-97 would be amended by inserting the word "the" in order to make clear the following term was directed to its antecedent. Support for this subject matter is as set forth in the previous version of these claims.

Claim 102 would be amended for purposes of clarity to recite "wherein the amino acid sequence of the human insulin precursor peptidyl fragment is the amino acid sequence of SEQ ID NO:4." Support for the subject matter of the amended claim is found *inter alia* in the previous version of the claim and in the first five lines at p. 25 of the specification.

Claim 103 would be amended for purposes of clarity to recite "wherein the amino acid sequence of the human insulin precursor peptidyl fragment is the amino acid sequence of SEQ ID NO:5." Support for the subject matter of the amended claim is found *inter alia* in the previous version of the claim and in the first five lines at p. 25 of the specification.

Claim 104 would be amended to recite "wherein the human insulin precursor peptidyl fragment consists of the A chain and B chain amino acid sequences of human insulin and therebetween a removable amino acid sequence of between 1 and 34 residues in length." Support for the "consists of" subject matter is found in the previous version of the claim which recited "comprising." Support for the removable subject matter is found *inter alia* in the specification at p. 24, first full paragraph, line 16.

Claim 113 would be amended to recite "wherein the amino acid sequence of the first peptidyl fragment is the amino acid sequence of SEQ ID NO:1." Support for the subject matter is as set forth in the specification at p. 16, lines 23-25.

Claim 114 would be amended to recite "wherein the amino acid sequence of the first peptidyl fragment is the amino acid sequence of SEQ ID NO:2." Support for the subject matter is set forth in the specification at p. 16, lines 26-29.

Claim 116 would be amended to recite "wherein the first peptidyl fragment is between 20 and 49 residues in length." Support for the subject matter of "49" is found *inter alia* in SEQ ID NO:1 at p. 35, which is 49 amino acid residues long.

Claim 122 was slightly reworded for purposes of clarity. Support is found *inter alia* in the previous version of the claim.

Claim 123 would be amended to recite in part:

> a first peptidyl fragment, wherein the amino acid sequence of the first peptidyl fragment is at least 60% identical to the amino acid sequence of at least the first 20 N-terminal amino acids of SEQ ID NO:1; and wherein the first peptidyl fragment is capable of being bound by an anti-human-growth hormone antibody;
>
> a human insulin precursor peptidyl fragment consisting of a human insulin precursor which exhibits insulin-like bioactivity when folded in a bioactive conformation; and wherein the human insulin precursor peptidyl fragment comprises the A chain and the B chain of human insulin, and wherein the A chain and the B chain are separated by a removable peptidyl moiety of between 1 and 34 residues in length, and

Support for the subject matter "SEQ ID NO:1" is found *inter alia* in the specification at p. 28 and original claim 4. Support for the anti-human growth hormone antibody subject matter is found in the specification at p. 16, third full paragraph. Support for the subject matter of "wherein the human insulin precursor peptidyl fragment comprises the A chain and the B chain of human insulin; and wherein the A chain and the B chain are separated by a removable peptidyl moiety of between 1 and 34 residues in length," is found *inter alia* in the specification in original claim 11 (2 to 34 amino acids in length) and SEQ ID NO:5 which provides an insulin precursor where the A and B chains are separated by one amino acid (arginine at position 31).

Claim 123 would also be amended to recite in part:

> wherein the first peptidyl fragment is capable of mediating, upon contacting of the chimeric protein with a chaotropic agent, the formation of a correctly folded conformation of the human insulin precursor peptidyl fragment.

Support for the above recital is found throughout the specification, see for instance the Abstract. Support for the "correctly folded" recital is set forth particularly in the specification at p. 24, second full paragraph.

Claim 124 would be amended to recite:

> The chimeric protein according to claim 123, wherein the amino acid sequence of the first peptidyl fragment is an amino acid sequence of SEQ ID NO:1 of the same length as the first peptidyl fragment.

Support for the above subject matter is found *inter alia* in the specification at p. 16, fourth full paragraph.

Claim 127 would be amended to depend from claim 124 and to recite "wherein the amino acid sequence of the human insulin precursor peptidyl fragment is the amino acid sequence of SEQ ID NO:4." Support for the above subject matter is found *inter alia* in the specification at p. 17, line 9.

Claim 128 would be amended to depend from claim 124 and to recite "wherein the amino acid sequence of the human insulin precursor peptidyl fragment is the amino acid sequence of SEQ ID NO:5. Support for the above subject matter is found *inter alia* in the specification at p. 17, line 17.

Claim 131 would be amended to recite in part:

> a first peptidyl fragment which has an amino acid sequence identical to the amino acid sequence of SEQ ID NO:1 through residues 1-20;
>
> a human insulin precursor peptidyl fragment comprising a human insulin precursor which exhibits insulin-like bioactivity when folded in a bioactive conformation; wherein the human insulin precursor peptidyl fragment comprises the A chain and the B chain amino acid sequences of human insulin separated by a removable peptidyl moiety between 1 and 34 residues in length, and
>
> an arginine or lysine residue or at least one cleavable peptidyl fragment linking the first peptidyl fragment and human insulin precursor peptidyl fragment;

Support for the above subject matter is as set forth for claims 78 and 123. Claim 131 would also be amended to recite "from contacting" in place of "upon contact of." Claim 131 would also be amended by adding indentations to simplify reading of the claim.

New claim 132 would depend from claim 78 and recite in part:

> wherein the recombinant protein consists of the first peptidyl fragment, the insulin precursor peptidyl fragment, and the linking peptidyl fragment, and wherein the first peptidyl fragment is identical in amino acid sequence to an amino acid sequence of SEQ ID NO:1 of at least 20 amino acids in length or a sequence variant thereof having only a small number of amino acid substitutions.

Support for the above recital is as set forth for claim 78. Support for the "small number" subject matter is found in the specification at p. 18, lines 20-25, which provides for a small number of base changes in a primer for expanding a nucleic acid encoding a subject chaperone polypeptide fragment in order to generate variants of chaperone polypeptides. One of ordinary skill would know that such base changes would provide variants of the chaperone polypeptide having a correspondingly small number of amino acids upon expression of the nucleic acid.

New claim 133 would depend from claim 132 and recite "wherein the small number is zero or one." Support is found *inter alia* at p. 22, line 24.

New Claim 134 would depend from claim 132 and recite "wherein the small number is two." The subject matter of "two" is inherently disclosed in a recital of "one or a small number." Such a small number must reasonably at least encompass the next higher small number of "two." Support is found *inter alia* at p. 22, line 24.

New claim 135 would depend from claim 132 and recite "wherein the small number is zero." When the small number is zero, the sequence has no substitutions. Therefore, the fragment comprises a sequence which is identical to that of SEQ of SEQ ID NO:1. Support for this subject matter is found *inter alia* in SEQ ID NO:1 at p. 35.

New claim 136 would depend from claim 78 and recite "wherein the human insulin A chain is identical in amino acid sequence to the amino acid sequence of residues 32-52 of SEQ ID NO:5 and the human B chain is identical in amino acid sequence to the amino acid sequence of residues 1-30 of SEQ ID NO:5." Support for this subject matter is found in the specification

at p. 17, paragraphs 3 and 4, p. 24, first full paragraph, at p. 36 and 37 (i.e., SEQ ID NO:4 and SEQ ID NO:5) which set forth the human A chain (residues 32-52 in SEQ ID NO:5 and residues 66-86 in SEQ ID NO:4) and human B chain (residues 1-30 in SEQ ID NO:5 or SEQ ID NO:4) separated by an arginine residue (residue no. 31 in SEQ ID NO:5 or residues 31-65 in SEQ ID NO:4). Figure 1A pertains to the human insulin precursor of SEQ ID NO:4 and Figure 1B sets forth the human A and B chain portions of mature human insulin.

New claim 137 would depend from claim 133 and recite "wherein the human insulin precursor peptidyl fragment consists of an amino acid sequence identical to the amino acid sequence of SEQ ID NO:4 or SEQ ID NO:5." Support is as set forth *inter alia* in the specification at p. 17, paragraphs 3 and 5.

New claim 138 would depend from claim 133 and recite "wherein the human insulin precursor peptidyl fragment consists of the amino acid sequences of the A chain and B chain amino acid sequences of human insulin and a removable amino acid sequence between 1 and 34 residues in length." Support for the above subject matter is found in the specification at p. 24, first full paragraph and as set forth for the insulin precursor subject matter of claim 123 above.

New claim 139 would depend from claim 104 and recite "wherein the recombinant protein consists of the first peptidyl fragment, the insulin precursor peptidyl fragment, and the at least one cleavable peptidyl fragment." Support for the above subject matter is found in the previous version of claim 104 and in specification at p. 16, first two paragraphs, and the paragraph bridging pp. 23 and 24.

New claim 140 would depend from claim 123, and recite "wherein the chimeric protein is identical in amino acid sequence to the amino acid sequence of SEQ ID NO:6 or of SEQ ID NO:7." Support for the above subject matter is found in the specification at p. 17, lines 18-22.

New claim 141 would depend from claim 124 and recite "wherein the human insulin A chain is identical in amino acid sequence to the amino acid sequence of SEQ ID NO:5 through residues 32-52 and the human B chain is identical in sequence to the amino acid sequence of SEQ ID NO:5 through residues 1-30." Support for the above subject matter is as described for claim 136 above.

New claim 142 would depend from claim 124 and recite "the chimeric protein of claim 124, wherein the protein consists of the first peptidyl fragment; the second peptidyl fragment; and an arginine or lysine residue or the at least one cleavable peptidyl fragment; and wherein the first peptidyl fragment is identical in amino acid sequence to an amino acid sequence of SEQ ID NO:1 of at least 20 amino acids in length, and wherein the at least one cleavable peptidyl fragment is at least 2 amino acids in length and has a C-terminal amino acid residue selected from the group consisting of Arg and Lys." Support for the above recital is as set forth for claim 123 and is found also *inter alia* in original claim 1.

New claim 143 would recite: "The chimeric protein of claim 124, wherein the first peptidyl fragment and the insulin precursor peptidyl fragment are linked by only one amino acid residue which is an arginine or lysine residue." Support for the above subject matter is found *inter alia* in original claim 1.

In view of the above, Applicants believe the above amendments to the claims add no new matter and respectfully request their entry.

VI. Responses to Claim Rejections Under 35 U.S.C. 112, 1st paragraph:

Standard of Review

Whether undue experimentation is required to practice an invention is typically determined by the Forman factors as set forth in MPEP §2164.01(a). These factors weigh (i) the relative skill of those in the art; (ii) the nature of the invention; (iii) the breadth of the claims; (iv) the amount of guidance presented; (v) the presence of working examples; (vi) the state of the art; (vii) the predictability of the art; and (viii) the quantity of experimentation necessary. *Ex parte Forman*, 230 U.S.P.Q. 546 (PTO Bd. Pat. App. & Inter. 1986), *In re Wands*, 858 F.2d 731, 8 U.S.P.Q.2d 1400 (Fed. Cir. 1988).

A. Response to the Rejection of Claims 78-97, 99, 102-104, 106-109, 111-114, 116, and 120-122 for an Alleged Lack of Enablement. (The similar rejections of claims 123 and 131 are addressed separately below.)

The Action rejected the above claims as allegedly not enabled particularly with respect to the scope of the first peptidyl or intramolecular chaperone portion of the protein and with respect to the scope of the second peptidyl fragment. Without acquiescing to the position of the Examiner and in order to expedite the prosecution of the application, Applicant has amended base claim 78 to recite in part:

> "a human insulin precursor peptidyl fragment comprising the human insulin A chain and the human insulin B chain;

The Applicant believes that the above amendment largely moots the rejection of the above claims based upon the breadth of the second peptidyl subject matter. Base claim 78 now is drawn in part to the A and B chains of human insulin. As noted in *Hormones* (1987), Chapter 7, (*see* IDS reference AK, pp. 286-289, and 278-284, and incorporated by reference), human insulin is one of the oldest studied human peptide hormones. Applicant trusts that the Examiner will recognize that the subject matter of such narrowly drawn human insulin precursor subject matter is well enabled. Applicant also notes that the recital of 'human' also largely moots any issue of antigenicity with respect to the second peptidyl fragment. SEQ ID NO:5 sets forth a sequence comprising the human A chain and B chain separated by an Arg residue at position 31.

Applicant hereinafter applies the above Foreman factor analysis to the first peptidyl fragment subject matter of claim 78 and its dependent claims.

1. *Breadth of the Claims*

Without acquiescing to the position of the Examiner, and in order to expedite prosecution of the application, the Applicant has amended the base claim to recite:

> 1) a first peptidyl fragment of about 20 amino acids in length to 92 amino acids in length having an amino acid sequence which is at least 60% identical to the sequence of SEQ ID NO:1 at least through residues 1 - 20 of SEQ ID NO:1 and wherein the first fragment is capable of being bound by an anti-hGH antibody,

[underlining added for emphasis].

The above recitals substantially further limit the size range of the first peptidyl fragment and the amount of amino acid sequence variation allowed. The amended claim requires a substantial homology to the indicated portion of the human growth hormone sequence. In particular, the recital of "wherein the first fragment is capable of being bound by an anti-hGH antibody," in particular, substantially changes the breadth of the first peptidyl subject matter. Antibody binding is highly dependent on the primary, secondary, and tertiary structure of a polypeptide and therefore greatly constrains the breadth of the "first peptidyl fragment" subject matter. As such, one of ordinary skill would appreciate that only a far less than an "infinite number" of modifications are contemplated.

In addition, claims 113, 114, 116, and 132-139 which depend directly or indirectly from base claim 78 set forth substantially narrower first peptidyl subject matter.

2. *The Predictability of the Art*

In assessing the predictability of the art, the Examiner cited some references which addressed the general prediction of protein structure and function from primary amino acid sequence information and some other references which addressed the effects of primary amino acid substitutions on the activity of the intramolecular chaperones. Applicant discusses the general and more specific references below.

i. Predicting Protein Function from Primary Sequence Information:

The Examiner cited Ngo, et al. (1994), the Wells (1990), Bork (2000), Doerks, et al. (1998), Smith, et al. (1997), Brenner (1999) and Skolnick, et al. (2000) as allegedly supporting the unpredictability of the art. The Ngo, et al. and the Skolnick, et al. references largely concern the *de novo* or *a priori* prediction of biological activity from the primary structure of a protein (i.e., the amino acid sequence). The Ngo, et al. and Skolnick, et al. references are not particularly on point as the claims read in part on variants of a first peptidyl fragment of specified sequence and activity. With respect to mutational variation of a known polypeptide of known function, the Wells reference is more on point in so far as it addresses the ability to predict *a priori* the effect of a given mutation or combination of mutations on the known

biological activity of a protein. However, as to all three of these references, enablement does not depend solely on making reliable *a priori* predictions. The standard of enablement is not one of *no* experimentation, the standard is one of no *undue* experimentation.[1] In this regard, Wells reports the effects of mutations to be generally additive and, more to the point, illustrates that in practice many such mutants - regardless of predicted activities - can be readily screened for their biological activity.

The Examiner also cited Bork (2000), Doerks, et al. (1998), Smith, et al. (1997), Brenner (1999) and Skolnick, et al. (2000) as allegedly supporting the unpredictability of the art. However, Applicant notes that the above references each concern the bioinformatics of predicting the function of an expressed nucleic acid sequence by analogy to other known nucleic acid sequences and their functions. Here, Applicants have set forth an amino acid sequence having a newly discovered useful activity and teach how to screen variants for the same activity. They are not charged with "discovering" the actual biological activity of an expressed sequence. Brenner (1999), Smith, et al. (1997), and Bork (1996) largely concern issues of annotation and genomic database management and quality control. These later three references are simply not relevant.

Bork, et al. (2000) is only slightly more on point and, to the limited extent of its relevance, does not support the Examiner's use of it. Table I of Bork reports selective examples of prediction accuracy for different areas of sequence analysis as ranging from 50% to 98%. Overall, Bork (2000) stands for the proposition that a 70% prediction is hard to achieve. However, a 70% accuracy rate does not indicate an undue amount of experimentation would be needed to find active variants. In fact, Bork (2000), to the limited extent it is relevant, indicates very little experimentation would be needed.

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[1] That some experimentation may be necessary to identify operative species does not constitute a lack of enablement. As the Federal Circuit has stated, "the key word is 'undue', not 'experimentation' " in determining whether pending claims are enabled. *Wands*, 8 U.S.P.Q.2d at 1405 (Fed. Cir. 1988). Indeed, a considerable amount of experimentation is permissible if it is merely routine, or if the specification in question provides a reasonable amount of guidance for practicing the invention.

ii. Predictability of the Intramolecular Chaperone Art.

Applicant notes that the references addressing the intramolecular chaperone art are more specific to the field of the invention and thus due much greater weight in assessing the predictability of the art.

Although cited for the opposite proposition by the Examiner, the Inouye, et al. (U.S. Patent No. 5,719,021) and Shinde and Inouye, et al. (1993) references actually support the claims as currently amended. As noted by the Examiner, the '021 patent teaches and claims the use of an exogenous activating polypeptide with a target polypeptide wherein the activating polypeptide has the sequence of the propeptide sequence of the target polypeptide or of a polypeptide with similar function and sequence. This patent issued with the following base claim:

> 15. An in vitro method to restore or increase the natural biological activity of a target polypeptide, which is normally expressed containing a prosequence, which target polypeptide is biologically inactive or has decreased natural biological activity due to improper folding of the polypeptide, which method comprises reacting intermolecularly in a buffered ionic aqueous medium, thereby favoring hydrophobic interaction, an exogenous activating peptide with the target polypeptide, wherein the activating peptide comprises an amino acid sequence that is substantially identical to a major portion of the amino acid sequence of the natural prosequence of the target polypeptide or of the prosequence of a polypeptide that is similar in amino acid sequence to the target polypeptide and has the same function as the target polypeptide, whereby the activating peptide promotes refolding the target polypeptide into its biologically active conformation.

The above claim is quite broadly drawn to, without *any* limitation as to target polypeptide or length of activating peptide, a 'chaperone' polypeptide subject matter in which the activating peptide comprises an amino acid sequence that is substantially identical to only a major portion of the amino acid sequence of the natural prosequence of the target polypeptide or of the prosequence of a polypeptide that is *similar* in amino acid sequence to the target polypeptide and has the same function as the target polypeptide, whereby the activating peptide promotes refolding the target polypeptide into its biologically active conformation. Applicant's claims, in contrast, are drawn to a much more narrowly drawn polypeptide target, comprising chain A and chain B of human insulin, and a human growth hormone chaperone which is, in particular,

tightly constrained by the limitation of being able to be bound by an anti-human growth hormone antibody.

The Shinde and Inouye, et al. (1993) reference was cited for the proposition that there are mutations which can eliminate or reduce the activity of a chaperone polypeptide. This reference gave emphasis to inactive mutants as they were assessing the important structural features of their chaperone polypeptide. The not unexpected existence of biologically *inactive* variants is simply not very determinative as to whether undue experimentation would be required to find *active* variants. Other results in Shinde and Inouye, et al. (1993) suggest not much experimentation would be required to find active variants. Table II of the Shinde and Inouye, et al. (1993) reference sets forth the conserved regions of the various polypeptide members of the subtilisin propeptide family. The conserved regions where sequence variation is not well tolerated comprise only very short portions of the total propeptide sequence. Indeed, U.S. Patent No. 5,719,021 to Inouye, et al. illustrates the relative ease with which a variety of mutants can be screened and tested for chaperone activity. Indeed, as noted above, claims to such broad subject matter were issued to Inouye, et al.

The Examiner notes that the specification states the results obtained with the subject human growth hormone related intramolecular chaperones was unexpected. However, the conclusion that the Examiner would draw from that fact is a non sequitur. While it was unexpected that a human growth hormone sequence could act as a intramolecular chaperone for an insulin precursor, **once that discovery was made**, one of ordinary skill in the art would subsequently expect that variants of the human growth hormone intramolecular chaperone polypeptide could readily be found which would work as well. Indeed, Inouye, et al., as discussed above, illustrates the approach where further variants are readily identified from the amino acid sequence once a chaperone polypeptide is known.

Applicant further calls the Examiner's attention to new dependent claims 132-135 and 137-138 which encompass a particularly narrow range of variation for the first peptidyl fragment subject matter.

3. *The State of the Art.*

With respect to the subject matter of intramolecular chaperones, the state of the art is sufficiently advanced to enable the several procedures necessary to practice the invention in the claimed scope. For instance, the Inouye, et al. patent teaches how to identify 'chaperone' polypeptides which are functional and similar in sequence to a known chaperone protein for a target polypeptide. It teaches that length and amino acid variations can be tolerated (*see* last full paragraph of col. 4 and next paragraph). Indeed, the reference expressly claims to be a general guide, it states at paragraph 2 at col. 7:

> As described hereinafter, the teachings of the invention are broadly applicable to any biochemically active mature polypeptide which is normally, but not necessarily, expressed in precursor form with a native pro-sequence from a biochemically inactive polypeptide which does not contain its pro-sequence. Illustrative types of such polypeptides, but not limited thereto include enzymes like various proteases, polypeptide, hormones, and other peptides which need a pro-sequence or its equivalent for folding into an active conformation. the claims

4. *Amount of Guidance Presented.*

The invention is based upon the surprising discovery that a human growth hormone polypeptide fragment could serve as an intramolecular chaperone. The state of the biotechnical arts is such that after that seminal teaching, the discovery of variants would be a matter of routine for one of ordinary skill in the biotechnical arts. Nevertheless, the specification teaches methods by which one of ordinary skill in the art can identify useful variants of the first peptidyl fragment. It teaches a first peptidyl fragment based upon the disclosed human growth hormone sequences (e.g., SEQ ID NO:1, 2) or variants thereof, it teaches methods for obtaining modified first peptidyl fragments with variations in their amino acid sequences and/or length, and teaches how to screen such fragments (p.28) for their chaperone activity; it also discloses features of the intramolecular chaperone fragment which are important to activity (e.g., relative percent of charged amino acids, polarized distribution) in the first paragraph at p. 13 and in the first paragraph at p. 25. The prior art already teaches the likely limited impact of conservative substitutions on protein function (see paragraph bridging cols. 9 and 10 of the '021 patent).

5. *Working Examples.*

The specification provides a working example starting at p. 29, wherein a first peptidyl fragment of about 45 residues in length is made and used to produce a properly folded insulin molecule.

6. *Relative Skill of those in the Art.*

As evidenced in the quality of the research and affiliations of the authors and inventors of the cited references, the level of skill of those in the art is high and fully commensurate with the skills needed to practice the instant claims.

7. *Nature of the Art.*

The art is in the field of recombinant protein expression and protein biochemistry. Both fields are advanced. The product of the methods, a properly folded insulin molecule, falls within a well developed field of pharmaceutics. In this field, a large amount of experimentation is routinely practiced.

8. *Amount of Experimentation Required.*

Variants of the first polypeptide sequence can be readily generated and tested as evidenced in the cited art and as instructed in the specification. Testing of such for chaperone activity can be readily carried out by *in vitro* techniques readily performed en masse by modern, analytical, and high through put methods well known to one of ordinary skill in the art.

Overall Summary of the Foreman Factors

Overall, the Forman analysis supports the enablement of the claimed subject matter:

1) The invention relates to the discovery that a human growth hormone sequence can serve as an intramolecular chaperone.

2) The claims have been amended to reduce the scope of the first peptidyl subject matter both as to sequence identity and length.

3) The level of skill in the protein/medicinal chemistry/intramolecular chaperone art is high.

4) The invention lies in the pharmaceutics/medicinal chemistry fields where much experimentation is expected and routine.

5) To the extent that the art is unpredictable, the cited evidence shows that undue experimentation would not be required to identify active first peptidyl fragments, and

6) The state of the art of intramolecular chaperones is fairly advanced and provides one of ordinary skill in the art with the tools needed to practice the invention.

7) The amount of guidance presented is commensurate with the claims and the teachings of the prior art.

8) The specification provides a working example illustrating the experimental manipulations needed to practice the claimed subject matter.

In view of the above, Applicant respectfully requests that the above rejections be reconsidered and withdrawn.

B. Response to the Rejection of Claims 123-129 for an Alleged Lack of Enablement

Claims 123-129 were rejected on grounds essentially the same as those described above. The above response thus is to be applied to the rejection of these claims as well. In addition, Claims 125, 126 and 129-130 have been canceled.

Without acquiescing to the position of the Examiner and in order to expedite prosecution of the application, Applicant would amend base claim 123 to recite:

> Claim 123: A chimeric protein comprising from N-terminus to C-terminus:
>
> a first peptidyl fragment, wherein the amino acid sequence of the first peptidyl fragment is at least 60% identical to the amino acid sequence of at least the first 20 N-terminal amino acids of SEQ ID NO:1; and wherein the first peptidyl fragment is capable of being bound by an anti-human-growth hormone antibody;
>
> a human insulin precursor peptidyl fragment consisting of a human insulin precursor which exhibits insulin-like bioactivity

> when folded in a bioactive conformation; and wherein the human insulin precursor peptidyl fragment comprises the A chain and the B chain of human insulin; and wherein the A chain and the B chain are separated by a removable peptidyl moiety of between 1 and 34 residues in length, and
>
> an arginine or lysine residue or at least one cleavable peptidyl fragment linking the first peptidyl fragment and the human insulin precursor peptidyl fragment;
>
> wherein the first peptidyl fragment is capable of mediating, upon contacting of the chimeric protein with a chaotropic agent, the formation of a correctly folded conformation of the human insulin precursor peptidyl fragment.

[underlining added for emphasis].

In light of the above amendments and previous remarks, Applicant submits that the first peptidyl subject matter is enabled.

Applicants note that claims 124, 127-128, and 140-143 are more narrowly drawn to first peptidyl subject matter wherein the first peptidyl fragment comprises an amino acid sequence identical to an amino acid sequence of SEQ ID NO:1. Applicants note that in paragraph 13, the Examiner stated that the subject matter of a first peptidyl fragment of SEQ ID NO:1 was enabled.

Applicant also notes that the Examiner had indicated that the subject matter of a second peptidyl fragment of SEQ ID NOs. 4 and 5 were enabled. The human A and human B chains of the insulin precursor comprise the corresponding A and B chains portions of SEQ ID NO:5, save the cleavable arginine at position 31 which may be subject to modification or elimination. Applicant notes that the cited reference U.S. Patent No. 4,916, 212 teaches and claims subject matter drawn in part to A chain and the B chain amino acid sequences of human insulin that are separated by an amino acid sequence between about 1 and 33 residues in length (see claim 1 of the '212 patent).

In view of the above, Applicant requests that the above rejections be reconsidered and withdrawn.

C. Response to the Rejection of Claim 131 for an Alleged Lack of Enablement

Claim 131 was rejected on grounds as set forth for base claim 78. Without acquiescing to the position of the Examiner and in order to expedite prosecution of the application, the Applicant would amend base claim 131 to recite:

> 131. A method of making a correctly folded human polypeptide with insulin bioactivity, said method comprising:
> expressing a recombinant protein comprising, from N-terminus to C-terminus:
> a first peptidyl fragment which has an amino acid sequence identical to the amino acid sequence of SEQ ID NO:1 through residues 1-20;
> a human insulin precursor peptidyl fragment comprising a human insulin precursor which exhibits insulin-like bioactivity when folded in a bioactive conformation; wherein the human insulin precursor peptidyl fragment comprises the A chain and the B chain amino acid sequences of human insulin separated by a removable peptidyl moiety between 1 and 34 residues in length, and
> an arginine or lysine residue or at least one cleavable peptidyl fragment linking the first peptidyl fragment and human insulin precursor peptidyl fragment;
> wherein the first peptidyl fragment is capable of increasing the yield of the bioactive conformation of the insulin precursor formed from contacting the protein with a chaotropic auxiliary agent as compared to the yield of the bioactive conformation of the insulin precursor formed from contacting the same protein lacking the first peptidyl fragment with the chaotropic agent;
> contacting the recombinant protein with an aqueous medium comprising the chaotropic agent; and
> cleaving at least one of the cleavable peptidyl fragments.

[underlining added for emphasis].

The above claim as amended recites a first peptidyl fragment "which has an amino acid sequence identical to the sequence of SEQ ID NO:1 through residues 1-20." This claim provides a human insulin precursor peptidyl fragment largely as set forth in claim 123. Thus, the above enablement responses with respect to the insulin precursor subject matter of base claim 78 and particularly base claim 123 also apply to base claim 131.

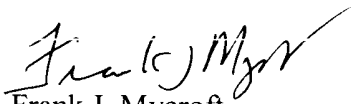
In view of the above, Applicant respectfully requests that the above rejection of claim 131 also be reconsidered and withdrawn.

CONCLUSION

In view of the foregoing, Applicant believes all claims now pending in this Application are in condition for allowance. The issuance of a formal Notice of Allowance at an early date is respectfully requested.

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 925-472-5000.

Respectfully submitted,



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APPENDIX A

ABSTRACT OF THE DISCLOSURE

The present invention relates to a chimeric protein containing an intramolecular chaperone (IMC) like sequence linked to a target protein, preferably an insulin precursor. The present invention also relates to a process for obtaining a correctly folded insulin-precursor-containing chimeric protein, comprising, *inter alia*, contacting an incorrectly folded chimeric protein containing an IMC like sequence linked to an insulin precursor with at least one chaotropic auxiliary agent. The present invention further relates to an assay for screening an amino acid sequence for the ability to improve folding of an insulin precursor using a chimeric protein containing an IMC like sequence linked to an insulin precursor.

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